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Symbiosis As The Way Of Eukaryotic Life: The Dependent Co-Origination Of The Body

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Molecular analyses of symbiotic relationships are challenging our biological definitions of individuality and supplanting them with a new notion of normal part–whole relationships. This new notion is that of a 'holobiont', a consortium of organisms that becomes a functionally integrated 'whole'. This holobiont includes the zoological organism (the 'animal') as well as its persistent microbial symbionts. This new individuality is seen on anatomical and physiological levels, where a diversity of symbionts form a new 'organ system' within the zoological organism and become integrated into its metabolism and development. Moreover, as in normal development, there are reciprocal interactions between the 'host' organism and its symbionts that alter gene expression in both sets of cells. The immune system, instead of being seen as functioning solely to keep microbes out of the body, is also found to develop, in part, in dialogue with symbionts. Moreover, the immune system is actively involved in the colonization of the zoological organism, functioning as a mechanism for integrating microbes into the animal-cell community. Symbionts have also been found to constitute a second mode of genetic inheritance, providing selectable genetic variation for natural selection. We develop, grow and evolve as multi-genomic consortia/teams/ecosystems.

1. Perceiving individuality

We are accustomed to thinking of animals as unique individuals. However, this is a matter of perception, not analysis. For biologists, individuality has been a major problematic concern. As EB Wilson noted in 1896,

> There is at present no biological question of greater moment than the means by which the individual cell-activities are coordinated, and the organic unity of the body maintained; for upon this question hangs not only the problem of the transmission of inherited characteristics, and the nature of development, but our conception of life itself.

Indeed 'our conception of life itself' is undergoing a remarkable revolution, as new technologies have enabled us to perceive not only 'entities' but also 'relationships' that would have otherwise been unnoticed. New technologies such as polymerase chain reaction (PCR), high-throughput RNA analysis, and next-generation sequencing have dramatically transformed our conceptions of the biosphere. They have revealed a microbial world of incredible complexity, and also a living world of complex and intermingled *relationships* – not only among microbes but also between microscopic and macroscopic life (Gordon 2012). Symbiosis is becoming one of the central principles of contemporary biology, replacing an essentialist conception of organismal 'individuality' with a new conception congruent with a larger systems biology approach. These discoveries have profoundly challenged the generally accepted view of 'individuals'.

In microbial and botanical sciences, symbiosis between a larger body and numerous smaller ones is taken as a matter of course. The acceptance of rhizobia, mycorrhizae and endocytic fungae as important components of plant life, for instance, is normative. Nonetheless, zoologists have subscribed to a more individualist conception of the organism,

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since the role of microbial symbionts had been more difficult to document in animal development, physiology and evolution (Sapp 1994, 2002, 2009). Zoological organisms have been defined as anatomical, embryological, physiological, immuno-logical, genetical or evolutionary individuals (Geddes and Mitchell 1911; Clarke 2010; Nyhart and Lidgard 2011; Gilbert *et al.* 2012). These conceptions, though, are not wholly independent of one another, and each of these definitions stems from the common tenet of genomic individuality: an individual comprises a single genome.

New data have revealed that animals are composites of many species living, developing and evolving together (Gilbert *et al.* 2012; McFall-Ngai *et al.* 2013). This discovery that symbiosis is the rule and not the exception in the animal kingdom is fundamentally transforming the classical conception of an insular individuality into one in which inter-active relationships among species creates a new realm of individuality, a 'holobiont' – the integrated organism resulting from host cells and persistent populations of symbionts. This notion challenges and seeks to replace the concept of a mono-genomic individual whose essential identity arises during development, is maintained by the immune system, and which is selected through evolution.

2.   Anatomical individuality

The *anatomical* individual animal is regarded as a structured whole. Yet, PCR data show that animals 'share' their bodies with numerous 'species' of bacteria and other microbes. The algal symbiont, *Symbiodinium*, provides most of the nutrients needed by its host coral (the term 'host' is used here in the classical sense to denote the larger, eukaryotic, multicellular organism in or on which the 'symbiont' resides). When this symbiosis is broken (for instance, by a prolonged increase in sea-surface temperatures), these corals 'bleach'. That is to say, they lose their algal symbionts and die. Similarly, the entity we call a cow is an organism whose complex ecosystems of gut symbionts – diverse communities of cellulose-digesting bac-teria, anaerobic fungi and ciliated protists – defines its plant-digesting physiology, and has played a determinative role in its evolution (Kamra 2005).

Mastotermes darwiniensis, a termite of northern Australia, is especially problematic in terms of individuality. The worker termites eat trees, digesting the cellulose in their guts and constructing elaborate subterranean nests. But the worker termite cannot digest cellulose without its gut symbiont, *Mixotricha paradoxa*, which is itself an anatomical composite of at least five other species, including a eukaryotic protest that has the nucleus, a bacterium that acts as a mitochondrion, about 250,000 *Trepinoma* spirochetes that provide locomotion, a large bacillus, and about 200 larger spirochetes. Margulis and Sagan (2001) aptly called it 'the beast with five genomes'.

It is estimated that nine out of every ten cells in the human body are bacterial (Bäckhed *et al.* 2005; Ley *et al.* 2006), and metagenomic sequencing (Qin *et al.* 2010) has shown that each human gut has entered into a persistent partnership with over 150 species of bacteria. The gene set contained by this symbiotic gut metagenome in the human species is about 150 times larger than that of the human eukaryotic genome. And this does not include the symbionts of human airways, skin, mouth, excretory or reproductive orifices.

Animals, therefore, cannot be regarded as individuals by anatomical criteria, but rather as holobionts, integrated organisms composed of both host cells and persistent popula-tions of symbionts (Rosenberg *et al.* 2007).

3.   Developmental individuality

The *developmental* view of animal individuality (Huxley 1852) is a variant of the anatomical version of biological individuality. Here, the individual animal is understood to be that which proceeds from ovum to ovum. This was critically important after Robert Remak and others showed that ani-mals were composed of myriads of smaller individuals, each alive in its own right (Nyhart and Lidgard 2011). Indeed, developmental mechanics (experimental embryology) cen-tered around the question of developmental individuality. Roux and Metchnikoff each had his own version respective version of Darwinian selection occurring within the embryo; and Driesch revolutionized embryology with his discovery that each cell of the early sea urchin embryo could produce a complete individual larva. How could a part become a whole? Spemann began his embryological studies with the observation that a partially constricted newt egg could give rise to a 'partially split individuality', having two 'egotisms' derived from a single egg. Somehow, the potentially inde-pendent and competitive parts of the embryo were normally integrated to form a singular individual whole (Hamburger 1988; Gilbert 1992).[1]

This notion of a dynamic part–whole relationship is being extended to symbionts as part of the development of the body. New evidence demonstrates that what we understand to be the 'individual' develops as consortia of animal cells and microbes (McFall-Ngai 2002; Gilbert and Epel 2009; Fraune and Bosch 2010; Pradeu 2011). Indeed, the develop-ment of both vertebrates and invertebrates (especially larval and postembryonic development) is predicated on intimate relations with microbes.

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[1] Even today, this ability of the whole to regulate its parts and to make the parts 'fit' is revelatory. Even some aggressive adult cancer cells, defined by their being 'autonomous', can become normally functioning parts of the body when placed into a particular portion of the embryo (Stewart and Mintz 1981; Kasemeier-Kulesa *et al.* 2008).

For instance, in numerous organisms, the development of particular organs depends on chemical signals from symbionts (Douglas 1988, 2010). The ovaries of the parasitoid wasp, *Asobara*, require signals from their *Wolbachia* symbionts if they are not to undergo apoptosis (Pannebakker *et al.* 2007). Newborns of the squid *Euprymna scolopes* lack a light organ. The instructions for making this organ are not encoded in the genome of the squid; rather, the squid embryo has evolved the ability to cooperate with one particular bacterial species, *Vibrio fischeri* (McFall-Ngai *et al.* 2012). Without this bacteria, the light organ does not develop. Without the squid, the luminescent genes of the bacteria are not expressed.

In mammals, the development of the immune and digestive systems is not completed without gut bacteria (Ley *et al.* 2006, 2008; Lee and Mazmanian 2010). 'Germ-free' (asymbiotic) mice have insufficient intestinal capillaries, poorly developed or absent gut-associated lymphoid tissue, and a T-cell repertoire so diminished that they have an immunodeficiency syndrome (Stappenbeck *et al.* 2002; Rhee *et al.* 2004; Niess *et al.* 2008; Duan *et al.* 2010). In zebrafish, microbes act through the canonical Wnt pathway to regulate the normal proliferation of the intestinal stem cells (Rawls *et al.* 2004; Bates *et al.* 2006). In both species, normal differentiation and growth of the gut depends on symbiotic microbes.

One particularly interesting area of microbial effects on holobiont development involves mammalian brain formation. Germ-free mice, for example, have lower levels of NGF-1A and BDNF (a transcription factor and a paracrine factor associated with neuronal plasticity) in relevant portions of their brains than do conventionally raised mice. Heijtz *et al.* (2011) concluded that 'during evolution, the colonization of gut microbiota has become integrated into the programming of brain development, affecting motor control and anxiety-like behavior'. Other investigators have noticed similar effects and have concluded that a 'microbe–gut–brain' axis exists (Rhee *et al.* 2009; Cryan and O'Mahony 2011; Collins *et al.* 2012; Cryan and Dinan 2012).

The relationship between symbiotic bacteria and the development of various cognitive states is now being explored (McLean *et al.* 2012; Mulle *et al.* 2013). The ingestion of certain *Lactobacillus* strains, for instance, lowers stress-induced corticosterone levels in mice and alters their behaviour. The behavioural effects correlated with the ability of this bacterial strain to increase the levels of the mRNA for a major GABA receptor subunit in the prefrontal cortex. Moreover, the effects of the bacteria can be abrogated by severing the region of the vagus nerve connecting the gut to the brain (Bravo *et al.* 2011). The ability of new gut microbes to help alleviate the anoxia of kwashiorkor similarly opens up an entire area of research (Smith *et al.* 2013).

Unlike insects, which can receive their microbial symbionts through the female germ cells, mammals acquire theirs by infection. As soon as the amnion breaks or when infants suckle or cuddle, microbes colonize the guts. Moreover, just as in traditional embryonic development, the microbes produce chemicals that induce appropriate gene expression in the neighbouring cells. In the developing guts of mice and zebrafish, hundreds of genes are activated by bacterial symbionts (Hooper *et al.* 2001; Rawls *et al.* 2004). In mice, the 'normal' levels of angiogenin-4 and colipase gene expression are those levels induced by the bacteria. The co-evolution of mammals and their gut bacteria has in effect resulted in the 'outsourcing' of developmental signals from animal cells to microbial symbionts. Mother's milk even contains sugars that the mammal cannot digest but which serve as food sources for the symbionts (Zivkovic *et al.* 2011). The symbionts are thus integrated into the normal networks of animal development, interacting with the eukaryotic cells of their 'host', and development is a matter of interspecies communication (Gilbert 2001, 2003; McFall-Ngai 2002). Development is a matter of interspecies communication. We not only co-evolve; we co-develop, and from the viewpoint of developmental biology we are not individuals.

4. Physiological individuality

The *physiological* view of animal individuality regards the organism as composed of parts that cooperate for the good of the whole (Milne-Edwards 1827; Leuckart 1851). The complexity of animal organization is seen to be accompanied by the increasing division of labour among organ systems, a concept analogous to Adam Smith's conception that socio-economic progress in complex societies results from the division of labour (Limoges 1994).

Molecular research has now demonstrated that symbionts can become part of an obligatorily integrated union (MacDonald *et al.* 2011; Vogel and Moran 2011). For example, the 'genome' of the mealy bug *Planococcus* is the product of a nested symbiosis: animal cells harbour the betaproteobacterium *Tremblaya princeps*, which in turn harbour a gammaproteobacterium, provisionally named *Moranella endobia*. The synthesis of amino acids appears to be coordinated between these two microbes and the host. Three of the enzymes needed for phenylalanine biosynthesis are encoded by the *Moranella* bacterium, five other enzymes are encoded by the *Tremblya* bacterium, and a final enzyme in this pathway is encoded by the genome of the insect itself (McCutcheon and von Dohlen 2011). Note that the genomes of *all three* organisms have been altered through this symbiosis. Such metagenomic sequencing has demonstrated the importance of microbes in insect physiological systems (Vásquez *et al.* 2012; Weiss *et al.* 2012).

Integrated host–symbiont biochemical pathways is characteristic of mammals as well; and a new concept, *cometabolism*,

has been introduced to describe the physiology of the holobiont (Smith *et al.* 2013). This notion reflects the findings that about one-third of a mammal's metabolome (the diversity of molecules carried in its blood) has a microbial origin (Wikoff *et al.* 2009; McFall-Ngai *et al.* 2013). The term was introduced to describe the findings that kwashiorkor was not just a disease of protein-poor diet. Rather, the disease originated through poor diet plus certain types of bacteria. The gut bacteria take our ingested foods and convert them into new products. What a person's cells experience is the result of cometabolism, 'a function of microbiota and host diet'.

Microbial symbiosis also has been demonstrated in vertebrate physiology. Specific bacteria induce the formation of regulatory T-lymphocytes that suppress potentially dangerous immune responses that can cause inflammatory bowel disease (Mazmanian *et al.* 2008; Chow *et al.* 2010). The role of symbiotic microbes in mammalian disease prevention is well recognized today (Mazmanian *et al.* 2008; Lee and Mazmanian 2010; Ballal *et al.* 2011), and different bacterial strains may alter how calories are obtained and may predispose mammals to weight gain and fat tissue formation (Ley *et al.* 2005; Turnbaugh and Gordon 2009; Everard *et al.* 2013). Indeed, bacteria may be critical in maintaining a woman's health during the last stages of pregnancy. When bacteria from pregnant women in their third trimester were transplanted into germ-free mice, the mice became fatter and developed insulin resistance, just as pregnant women do. This did not happen with the bacteria from first-trimester pregnant women (Koren *et al.* 2012). Microbial symbionts appear to be a normal part of animal physiology, working toward a functional holobiont. We are not individuals by physiological criteria.

5. Genetic individuality

Microbial symbionts form a second type of genetic inheritance (Moran 2007; Gilbert 2011). Arthropods often acquire their symbionts vertically though the maternal germline as well as horizontally from the environment, while mammals usually obtain them from the maternal reproductive tract and from the interactions following birth. In aphids, symbiotic bacteria provide selectable allelic variation (such as thermotolerance, colour and parasitoid resistance) that enable some holobionts to persist better under different environmental conditions (Dunbar *et al.* 2007; Tsuchida *et al.* 2010). Thus, whether the holobiont can reproduce in hot weather, have cryptic coloration or survive a parisitoid wasp infection depends not on 'its' genome but on the genome of its symbionts. There is also allelic variation in the human microbiome. The genes of *Bacteroides plebeius* differ in different human populations. The Japanese strain contains at least two genes (horizontally transferred from a marine relative) that

enable the bacteria to metabolize complex sugars, such as those found in seaweeds (Hehemann *et al.* 2010, 2012). Indeed, the Human Microbiome Project (Turnbaugh *et al.* 2007) has applied ecological metagenomics to explore the microbial world within the human species.

In some species, the symbiont will actually bias its transmission. The common arthropod symbiont, *Wolbachia*, is only transmitted through the female germline (i.e. by the oocyte.) Males do not transmit these bacteria. In *Amadillidium* (the pillbug), *Wolbachia* will override the host's genetic mechanisms of sex determination, changing the development of the organism from male to female (Cordaux *et al.* 2004).

Symbionts can give us selectable genetic variation and comprise a second path of genetic inheritance. Our bodies have multiple genotypes.

6. Immunological individuality

The discipline of immunology has been called 'the science of self/non-self discrimination' (Klein 1982). In this view, the immune system consists of defensive 'weaponry', evolved to protect the body against threats from pathogenic microbes. Accordingly, if it were not for the immune system, opportunistic infections would prevail (as they do in cases of immune deficiencies), and the organism would perish.

In a fascinating inversion of this view of life, recent studies have shown that an individual's immune system is in part created by the newly acquired microbiome. In vertebrates, the gut-associated lymphoid tissue is specified and organized by bacterial symbionts (Rhee *et al.* 2004; Lanning *et al.* 2005). When symbiotic microbes are absent in the gut, the immune system fails to function properly and its repertoire is significantly reduced (Lee and Mazmanian 2010; Round *et al.* 2010). Similarly, Hill *et al.* (2012) have shown that microbial symbionts provide developmental signals that limit the proliferation of basophil progenitor cells and thereby prevent basophil-induced allergic responses. Lee and Mazmanian (2010) conclude, 'multiple populations of intestinal immune cells require the microbiota for their development and function'.

The immune system, therefore, appears to be more of a 'passport control agent' or even a 'bouncer' rather than a defensive army posted to keep the zoological organism 'pure'. It distinguishes, by evolutionary experience, between potential symbionts and the potential pathogens (Matzinger 1994). Indeed, the immune system actively recruits the symbionts. Peterson *et al.* (2007) have shown that intestinal IgA, in addition to its well-known role in attacking pathogens, plays a 'critical role in establishing a sustainable host-microbial relationship'. Similarly, these Peyer's patch antibodies, which are essential in fighting opportunistic pathogens, appear to be

involved in 'the creation of an optimal symbiotic environment on the interior of the PPs' (Obata *et al.* 2010). Even the Toll-like receptors that mediate innate immunity are utilized by *Bacteroides* to establish a host–commensal relationship. The ability of symbiotic bacteria to use the innate and acquired immunity pathways to initiate symbioses has led Round *et al.* (2011, p 974) to conclude that 'the immune system can discriminate between pathogens and the microbiota through recognition of symbiotic bacterial molecules in a process that engenders commensal colonization'. In squids (McFall-Ngai *et al.* 2010) and mammals (Hooper *et al.* 2012), elements of the host immune system have been co-opted to support the colonization, limitation and persistence of symbiotic bacteria within the host.

Thus, the immune system, built, in part, under the supervision of microbes, does not merely guard the body against other hostile organisms in the environment but it also mediates the body's participation in a community of 'others' that contribute to its welfare (Tauber 2000, 2008; Agrawal 2001; Hooper *et al.* 2001; Dale and Moran 2006). What counts as an individual is now seen as being dynamic, context-dependent and responsive to symbionts.

7. Evolutionary individuality: The revised individual

Biological individuality has also been defined evolutionarily, as that which can be selected. The individuals are usually considered to be genes or monogenomic organisms. But, from the above discussion, it is evident that organisms are anatomically, physiologically, developmentally, genetically and immunologically multigenomic and multispecies complexes. Can it be that organisms are *selected* as multigenomic associations? Is the fittest in life's struggle the multispecies consortium, and not an individual of a single species in that group? This possibility has been raised by Bateson (1988), who argued that 'the outcome of the joint action of individuals could become a character in its own right'.

An instructive example comes from studies of the pea aphid, *Acyrthosiphon pisum*, and the several species of bacteria that live within its cells. Variants of *Buchnera* spp. bacteria provide the aphid with thermotolerance (at the expense of fecundity at normal temperatures; Dunbar *et al.* 2007); *Rickettsiella* bacteria provides a pathway for aphid colour change, turning genetically red aphids green through the synthesis of quinones (Tsuchida *et al.* 2010); and some variants of the bacteria *Hamiltonella* spp. provide immunity against parasitoid wasp infection (Oliver *et al.* 2009). However, in the last case, the protective variants of *Hamiltonella* result from the incorporation of a specific lysogenic bacteriophage within the bacterial genome. The aphid must be infected with *Hamiltonella*, and the *Hamiltonella* must be infected by phage APSE-3. As Oliver *et al.* (2009, p 994) write, 'In our system,

the evolutionary interests of phages, bacterial symbionts, and aphids are all aligned against the parasitoid that threatens them all. The phage is implicated in conferring protection to the aphid and thus contributes to the spread and maintenance of *H. defensa* in natural *A. pisum* populations'. But there is a trade-off to the host in having this beneficial protection. In the absence of parasitoid infection, those aphids carrying the bacteria with lysogenic phage are not as fecund as those lacking this phage. A similar trade-off occurs in aphids that carry the thermotolerant alleles of *Buchnera*. Those aphids whose symbionts bear the heat-resistant allele have less fecundity at milder temperatures than their sisters whose bacteria lack the functional allele for the heat-shock protein. However, the *population* as a whole can survive hot weather, which would otherwise prevent reproduction. As Bateson (1988) had pointed out, it is the whole that gets selected, and that 'focusing on the genetics of individuals muddles the issue of what is necessary for differential survival with what is required for replication'.

But if animals are not genetic or anatomical individuals, and if there is no 'individual organism', what remains of classic notions of 'individual selection'? This moves the biological discussion of symbiotic associations into the arena of 'group selection'. Most discussions of group selection, however, are not germane here, because they assume that the group in question is composed of members of a single species. However, one important concern is relevant to our discussion of the holobiont: cheaters. The major problem for all group selection theories (and the groups themselves) are potential 'cheaters', those lower-level parts of the group that would proclaim their own autonomy and that would multiply at the expense of the others. The problem of 'cheaters' then has to be solved in such a way that associates in a symbiotic relationship are under the social control of the whole, the holobiont (Stearns 2007).

This strong socializing and unifying force is found in the immune system, and here we find a solution to the problem of cheaters in a symbiotic complex. As mentioned earlier, the immune system must now be formulated as having two 'limbs': an *outward*-looking limb that defines the organism as that which is to be protected from foreign pathogens, and an *inward*-looking arm that looks for potential dangers arising from within the organism itself (Burnet and Fenner 1949; Tauber 2000, 2009; Ulvestad 2007; Eberl 2010; Pradeu 2010). This dualistic vision was the original conception of Metchnikoff at the end of the 19th century (Tauber 1994). If the immune system serves as the integrating system, keeping the animal and microbial cells together, then to obey the immune system is to become a citizen of the holobiont. To escape immune control is to become a pathogen or a cancer. Cheaters are destroyed by the immune system.

Thus, the symbionts are welcomed into the animal body and are regulated by the immune system. As part of the

body, the microbes not only help the body develop and remain physiologically intact, they also provide a secondary system of genetic transmission from parent to offspring. They can provide selectable variation from generation to generation. Moreover, in addition to providing this selectable variation, microbial symbionts may have played, and continue to play, other roles in animal evolution as well (Margulis and Fester 1991). Animal speciation may be mediated, in part, through the ability of microbes to induce reproductive isolation. This can be achieved through symbiont-induced cytoplasmic incompatibility between hybrids (Bruckner and Bordenstein 2012) or by symbiont-induced mate selection (Sharon *et al.* 2010). In many arthropods, those animals infected with one strain of *Wolbachia* cannot mate with members infected with a different strain. *Wolbachia* appears to be producing a reproductive barrier separating species into two groups. In the wasp *Nasonia*, *Wolbachia*-induced cytoplasmic incompatability appears to be the event that split a group of such wasps into *Nasonia giraulti* and *Nasonia longicornis* (Bordenstein *et al.* 2001). Here, the wasps can interbreed if the symbionts are removed; but the hybrids between them die if the symbionts are present.

Reproductive isolation can also be achieved by influencing mate selection. This appears to be the case in *Drosophila,* where mate selection has been shown to be dependent on symbionts that are introduced into the fly larvae through its food. If the symbionts are removed, the mating preference is abrogated (Sharon *et al.* 2010). In both these cases, symbionts effect reproductive isolation, a critical *sine qua non* of speciation.

Last, we may never have been 'pure' animals, innocent of symbionts. We must remember that eukaryotic protists were not only created by endosymbiosis, but that the protist world is full of complex symbioses (Margulis 1981; Margulis and Fester 1991; Sapp 2009). Some of these microbial symbioses may have led to multicellularity. The choanoflagellates are unicellular protists that are the sister group of the animals. However, these unicellular forms can be converted into multicellular entities – complete with an extracellular matrix and cytoplasmic bridges between cells – by a specific bacterium that often coexists with them. If the protists are cultivated in filtered water, they remain unicellular. If the bacteria are added back, they can form multicellular entities (Dayel *et al.* 2011; Alegado *et al.* 2012). Thus, bacteria symbionts may have been important in initiating multicellularity, the ultimate part/whole dialectic in biology.

Animals, then, can no longer be considered individuals in any sense of classical biology: anatomical, developmental, physiological, immunological, genetic or evolutionary. Our bodies must be understood as holobionts whose anatomical, physiological, immunological and developmental functions evolved in shared relationships of different species (Zilber-Rosenberg and Rosenberg 2008). Thus, the holobiont, with its integrated community of species, becomes a unit of natural selection whose evolutionary mechanisms are largely unexplored (Savinov 2011). Indeed, if the incorporation of new participants, with novel genetic and physiological properties, into evolving holobionts has been a common evolutionary strategy, some major tenets of the Modern Synthesis may need re-evaluation. As Lewis Thomas (1974, p142) commented when considering self and symbiosis: 'This is, when you think about it, really amazing. The whole dear notion of one's own Self – marvelous, old free-willed, free-enterprising, autonomous, independent island of a Self – is a myth'. Our new biology will have to deal with this.

Acknowledgements

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